

**In the Specification:**

[0065] Fig. 3A, Fig. 3B and Fig. 3C are photomicrographs (magnification: times 40; bar length on photograph = 2mm) of the root and root hairs of representative five day old seedlings obtained from representative WT, AVP1 transgenic and AVP2 transgenic of Fig. 3A grown parallel to the surface on vertical plant nutrient agar plates.

[0068] Fig. 5 is a perspective view of wild type plants (WT) versus representative transgenic plants overexpressing AVP1 (AVP1-1 and AVP1-2) grown in salty soil. Five wild-type plants (WT) and two AVP-1 overexpressing transgenic lines (AVP1-1 and AVP1-2) were grown on soil in a 10 hour light/dark cycle. Plants were watered with a diluted nutrient solution (1/8 MS salts) for six weeks and subsequently watered with a diluted nutrient solution supplemented with NaCl. The concentration of NaCl began with 100 mM and was increased every four days by 100 mM. The photograph in Fig. 3 Fig. 1A corresponds to plants at the tenth day in the presence of 300 mM NaCl. Fig. 3 illustrates that the two AVP-1 plant types (aVP1-1 and AVP1-2) were significantly harder in salty soil as compared to wild-type plants. The fact that genetically engineered *Arabidopsis thaliana* plants that overexpress either AVP1 (the pyrophosphate-energized vacuolar membrane proton pump, this work) or AtNHX1 (the  $\text{Na}^+/\text{H}^+$  antiporters, (Apse, M., et al., *Science*, 285:1256-1258 (1999)) and this work) are capable of growing in the presence of high NaCl concentrations strongly supports the strategy described herein. A double transgenic plant would be expected to demonstrate a further enhanced salt tolerant phenotype. These *Arabidopsis thaliana* transporters or their counterparts may perform similar function in important agricultural crops. Fig. 5 is a drawing of wild type plants (WT) versus representative transgenic plants overexpressing AVP1 (AVP1-1 and AVP1-2) grown in salty soil.

[0072] Two transgenic lines of *Arabidopsis thaliana* were analyzed, AVP1-1 and AVP1-2. Each line contains extra copies of the 35S:AVP1 gene inserted at a single chromosomal location. Analysis of AVP1 protein levels in membrane fractions isolated from shoots show shows that these transgenic plants express more AVP1 protein than does the wild type (AVP1- 1, 1.6 fold and AVP1- 2, 2.4 fold increase over wild type, P-value = 0.0005) (Fig. 1) (Fig. 4) as determined from four independent immunoblots Western blots. The differences between these transgenic plants could be due to the number of copies of AVP1 inserted into the genome or the sites of insertion.

[0073] The enhanced tolerance to salinity and drought in transgenic plants with increased levels of AVP1 is most easily explained by an enhanced uptake of toxic cations such as sodium into the vacuole. Presumably, the greater AVP1 activity provides increased  $\text{H}^+$  to drive the secondary active uptake of cations into the lumen of the vacuole (Fig. 2C 6C).

[0074] Fig. 6 6A and 6B show a graph of  $\text{Na}^+$  and  $\text{K}^+$  content of wild-type plants (WT) versus...

[0076] The above data are is consistent...

[0077] It is well documented that  $\text{Ca}^{++}$  enters the plant vacuole via a  $\text{Ca}^{++}/\text{H}^{+}$  antiporter....Fig. 8 Fig. 7 shows that  $\text{Ca}^{++}$  uptake in ...

[0080] Transgenic plants...Referring to Fig. 1, Fig. 9, ...

[0081] To demonstrate...Referring to Figs 2A and 2B, Figs 10A and 10B, ...Referring again to Figs. 2A and 2B, Figs. 10A and 10B, ...

[0083] Similar results...The volume of five seed pods from the crossed lines of tobacco plants remained between about 1.2 milliliters and 1.4 milliliters (see Fig. 11).

**In the Claims:**

Please cancel claims 38-40, 43-45, 47-50, 52-53, 56-57, 59-65, 68-69, 71-76 and 78-83.

Please amend claims 34, 35, 36, 41, 51, 58, 66, 70 and 77.

Claims 84-104 have been added.

Claims 34-37, 41-42, 51, 54-55, 58, 66-67, 70, 77 and 84-104 are presented in the case.

**REMARKS**

**Objections to the Specification:**

[0065]: We have studied paragraph [0065] of the specification and do not believe that text is missing; however, applicants respectfully request additional clarification should the examiner disagree; however, Applicants have included this part of section [0065] in this response for the examiner's convenience in comparing documents. Applicants solicit examiner's further clarification in this regard.

[0068]: The examiner has noted what appear to be inconsistencies in reference to the figures in the specification. Paragraph [0068] refers to figure 5, then to figure 3, which on the face appears confusing. Applicants agree and have carefully reviewed the application for consistency in references to the figures.

Applicants have amended paragraph 0068 to conform to the specification as follows:

Fig. 5 is a drawing view of wild type plants (WT) versus representative transgenic plants overexpressing AVP1 (AVP1-1 and AVP1-2) grown in salty soil.